REMARKS

Claims 117-120 and 124-132 are pending in this application. Claims 130-132 have been withdrawn from prosecution as directed to non-elected subject matter. Claim 121-124 and 133-135 have been previously cancelled.

Claims 117, 118 and 127-129 have been objected to, and claims 117-120 and 124-129 have been rejected under 35 U.S.C. §§ 112, second paragraph, and 103(a). In view of the amendments to the claims and remarks below, Applicants respectfully request that the objections and rejections be reconsidered and withdrawn.

OBJECTION TO THE CLAIMS

Claims 117, 118 and 127-129 have been objected to for containing certain informalities. Applicants have amended these claims in accordance with the suggestions made by the Examiner, or have deleted the portions of the claims containing an informality. Accordingly, withdrawal of these objections is respectfully requested.

REJECTION UNDER 35 U.S.C. § 112, SECOND PARAGRAPH

Claims 117-120 and 124-129 have been rejected under 35 U.S.C. § 112, second paragraph, as being indefinite because claims 117 and 124 recite certain relative terms or exemplary phrases. Applicants have deleted these relative terms and exemplary phrases from claims 117 and 124.

These claims were further rejected because "it cannot be determined from the language set forth in step (e) of independent claim 117 when washing the pellet obtained in step (d) is required."[1] This optional step has been deleted from step (e).

These claims were also rejected "because it is unclear what is intended by the recitation 'the processed sample being used in the form of smear, culture, or Polymerase chain reaction (PCR) starting material, using PCR amplifiable mycobacterial DNA, and RNA.'"[2] This portion of claim 117 has also been deleted.

[1] Office Action at page 4.

[2] Office Action at page 4.

Claim 125 has been rejected as indefinite for its recitation of "wherein said method is culture runs at a neutral pH." Claims 125 has been amended to recite "wherein said mixing, homogenizing, adding, first washing, second washing, and resuspending steps are performed ~~method in culture runs~~ at a neutral pH."

Claim 127 has been rejected as indefinite because it appears to be incomplete. Applicants have added the step suggested by the Examiner.

Claims 128 and 129 have been rejected as indefinite because they recite "the primers", which lacks antecedent bases. Applicants have amended these claims appropriately.

Accordingly, withdrawal of these rejections is respectfully requested.

REJECTION UNDER 35 U.S.C. § 103

Claims 117-120 and 124-129 have been rejected under 35 U.S.C. § 103 in view of the following combination of references:

Claim(s)	Cited References
117-120 and 124-126	Chakravorty[3], Jaber[4] and Hernstadt[5]
127-129	Chakravorty, Jaber, Hernstadt, GenBank[6], Marchetti[7] and Buck[8]

Applicants respectfully traverse these rejections for the reasons set forth below.

I. REJECTION OF CLAIMS 117-120 AND 124-126

This invention, as recited in amended claim 117, a method of processing clinical samples useful for diagnosis of bacterial infections. The method recites six solutions:

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[3] Chakravorty *et al.*, "Novel use of guanidinium isothiocyanate in the isolation of *Mycobacterium tuberculosis* DNA from clinical material," FEMS MICROBIOLOGY LETTERS (2001) 205: 113-117 ("Chakravorty).

[4] Jaber *et al.*, "A simple method of DNA extraction from *Mycobacterium tuberculosis*," TUBERCLE AND LUNG DISEASE (1995) 76: 578-581 ("Jaber").

[5] United States Patent No. 6,027,883 to Herrnstadt *et al.* ("Herrnstadt").

[6] GenBank Accession No. U22037 ("GenBank").

[7] Marchetti *et al.*, "Evaluation of PCR in detection of *Mycobacterium tuberculosis* from formalin-fixed, paraffin-embedded tissues: comparison of four amplification assays," J. OF CLINICAL MICROBIOLOGY (1998) 36(6): 1512-1517 ("Marchetti").

[8] Buck *et al.*, ""Design strategies and performance of custom DNA sequencing primers," BIOTECHNIQUES (1999) 27(3): 528-536 ("Buck")

- Solution 1: a Universal Sample Processing (USP) solution comprising 3-6 M Guanidinium Hydrochloride (GuHC1), 40-60 mM Tris-Cl at a pH ranging between 7.3-7.7, 20-30 mM EDTA, 0.3-0.8% Sarcosyl, and 0.1-0.3 M beta-mercaptoethanol;
- Solution 2: 65 to 70 mM sodium phosphate at pH ranging between 6.7 to 6.8, or sterile water;
- Solution 3: 0.03 to 0.08% of polysorbate 80;
- Solution A: 8-12% a chelating resin;
- Solution B: 0.02 to 0.04% polyoxyethylene phenyl ether; and
- Solution C: 0.2-0.4% polysorbate 20

The method comprises obtaining the clinical sample. The clinical sample is mixed with 1.5 to 2 volumes of Solution 1. The clinical sample and Solution 1 are homogenized in a manner that avoids frothing. Solution 2 is added to the homogenate followed by centrifugation to obtain a pellet. The pellet is washed with Solution 1 and then optionally washed with water. The pellet is respsended in one or more of Solutions 3, A, B and/or C to obtain a processed sample.

Chakravorty discusses a DNA isolation method.[9] The method consists of homogenizing a tissue in 5M GITC, 50mM Tris-CL, pH 7.5, 25 mM EDTA 0.5% Sarcosyl, 0.2 M β-mercaptoethanol, which Chakravorty refers to as an "inhibitor removal solution" or "IRS".[10] As the Examiner acknowledges, Chakrovarty does not teach using GuHCl.

The Examiner contends that Chakrovarty section 2.2.1 adding sterile water to the homogenate because Chakrovarty's IRS inherently includes water. Assuming that Chakrovarty's IRS inherently includes water, Chakrovarty does not separately teach to add water to a homogenate. At best, it teaches adding IRS and water to a clinical sample, not a homogenate.

[9] Chakravorty at page 114.

[10] *Id.*

According to Charkovarty's method, once homogenized, the sample is centrifuged and the supernatant discarded.[11] The pellet is resuspended in the IRS, centrifuged, and the subsequent pellet is rinsed with water and dried.[12]

Jaber is directed to a method of DNA extraction of *M. tuberculosis*.[13] Jaber's method consists of incubating a mycobacterium culture in a lysis buffer consisting of 6M guanidinium HCl, 50mM EDTA, 1 mM 2-mercaptoethanol and 0.05% Tween 80.[14] After the bacteria is incubated in the lysis buffer, the sample is centrifuge. The resulting supernatant transferred into a clean tube, and the DNA precipitated with cold ethanol.[15]

Applicants respectfully disagree that Chakoravorty teaches adding solution 2 to the homogenate, or that one would find it obvious to substitute GITC with GuHCl in view of these references. Taking Chakorvaorty as a whole, one would not reasonably conclude that solution 2 is added to the homogenate. Furthermore, the Examiner contends that Jaber teaches that "GuHCl was a chaotropic agent that use useful for cell lysis, inactivation of nucleases, dissociation of nucleoproteins and disturbance of cellular and subcellular structures (see page 579)."[16] Applicants respectfully disagree because Jaber teaches that GuHCl is useful in lysising mycobacterium, not tissue; and because the Examiner has not explained why one would simply substitute Charkovarty's GITC with Jaber's GuHCl as opposed to substituting Charkovarty's lysis buffer for Jaber's lysis buffer.

Point I. Chakrovarty section 2.2.1 does not teach a separate step of adding solution 2 to a homogenate.

When making a rejection under 35 U.S.C. § 103, the examiner has the burden of establishing a *prima facie* case of obviousness. *In re Fritch*, 23 U.S.P.Q.2d 1780, 1783 (Fed. Cir. 1992). To establish this, each and every claimed element must be taught or made obvious by the applied references. *Ex parte Hellums*, Application No. 09/103,704, Appeal

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[11] *Id.*

[12] *Id.*

[13] Jaber at 578.

[14] *Id.* at 579.

[15] *Id.*

[16] Office Action at page 12.

No. 2001-2694, 2003 WL 25281923 at *4 (BPAI Jul. 15, 2003); *Ex parte Likins*, Application No. 10/010,392, Appeal No. 2004-0760, 2004 WL 4981756 at *3 (BPAI Apr. 8, 2004).

The Examiner contends that Chakrovarty teaches adding a second solution to the homogenate because solution 1 inhererntly includes water by citing Chakrovarty section 2.2.1. This section of Chakrovarty states:

> Mince and homogenize tissue in 5 M GITC, 50mM Tris-Cl, pH 7.5, 25 mM EDTA, 0.5% Sarcosyl, 0.2 M β-mercaptoethanol (inhibitor removale solution, IRS) in a mini bead beater (Biospec, USA) using 1-mm glass beads for 30-60 s. Centrifuge at 600 x g for 3 min. Centrifuge the supernatant at high speed and discard supernatant.

In contrast, claim 117 recites "... mixing 1.5 to 2 volumes of Solution 1 with the sample, homogenizing the mixing while avoiding frothing, adding Solution 2 to the homogenate followed by centrifugation to obtain a pellet ...." Thus, it recites that solution 1 is added to the sample. The sample and solution 1 are homogenized. After the sample is homogenized, solution 2 is added to the homogenate. Even assuming that a fair reading of Chakrovarty would include a teaching of solution 2 (i.e. sterile water), it does not teach adding sterile water in a separate step.

Moreover, a fair reading of Chakrovarty does not include that sterile water, instead of IRS, can be used. To establish a *prima facie* case of obviousness, the prior art must be evaluated based on what it, *as a whole*, teaches to one of ordinary skill in the art. *In re McLaughlin*, 443 F.2d 1392 (CCPA 1971). Here, Chakrovarty, as a whole, teaches that the IRS is not sterile water. Even assuming that IRS comprises sterile water, for the purposes for a rejection under Section 103, a fair reading of Chakrovarty, as a whole, would not prompt one of ordinary skill to conclude that sterile water can be used instead of Chakrovarty's IRS. Nor would one of ordinary skill in the art reasonably expect that Chakrovarty's DNA isolation method would work if Chakrovarty's IRS was substituted with sterile water.

A. Response to Office Action of April 8, 2009

The Examiner contends that Chakravorty's IRS solution contains sterile water. Assuming this is true, this does not teach adding sterile water to the homogenate. It teaches adding sterile water to a clinical sample before homogenization.

Accordingly, Chakrovarty does not suggest adding solution 2 to a homogenate, nor does it suggest solution 2 in and of itself.

Point II. The Examiner has not provided a reason why one would substitute Chakrovarty's GITC for Jaber's GuHCl when Jaber only teaches lysing mycobacterium, not tissue.

There is no reason to expect that Chakrovarty's GITC can be substituted with Jaber's GuHCl when isolating DNA from tissue according to Chakrovarty's method because Jaber's GuHCl was only disclosed as being useful in isolating DNA from bacteria, which requires less stringent conditions that isolating DNA from tissue. When making a rejection under 35 U.S.C. § 103, the Examiner has the burden of establishing a *prima facie* case of obviousness. *In re Fritch*, 23 U.S.P.Q.2d 1780, 1783 (Fed. Cir. 1992). To establish a *prima facie* case of obviousness, the prior art must be evaluated based on what it, as a whole, teaches to one of ordinary skill in the art. *In re McLaughlin*, 443 F.2d 1392 (CCPA 1971).

As part of a *prima facie* case, an examiner must establish some reason to combine the references. *KSR Int'l Co. v. Teleflex Inc.*, 127 S.Ct. 1727, 1731 (2007); *Takeda Chemical Industries, Ltd. v. Alpharpharm Pty., Ltd.*, 492 F.3d 1350, 1356-1357 (Fed. Cir. 2007). The *KSR Int'l* Court acknowledged the importance of identifying a reason that would have prompted a person of ordinary skill in the art to combine the elements in the way the claimed invention does. *KSR Int'l*, 127 S.Ct. at 1731; *Takeda Chemical*, 492 F.3d at 1356-1357. Repeatedly throughout the *KSR Int'l* decision, the Court discussed the importance that the result obtained by a particular combination was predictable to one of ordinary skill in the art. *KSR Int'l*, 127 S.Ct. at 1731 and 1739-1742.

Chakrovarty teaches lysing tissue with a lysis buffer comprising GITC, whereas Jaber teaches lysing a bacteria with GuHCl. There are also more differences than just GITC and GuHCl between Chakrovarty's and Jaber's lysis buffers. There is no explanation why one would reasonably expect GuHCl to lyse a tissue when Jaber only teaches lysing bacteria, and there is no explanation why one would reasonably expect GuHCl to work in Chakrovarty's formulation.

Jaber discloses to a protocol using GuHCl for extracting DNA from mycobacterium. In contrast, Chakravorty discloses a protocol using GITC for extracting

DNA from tissue. Thus, in order for the references to be properly combined, there must be some reason why one would reasonably expect that GuHCl would be equally as useful in extracting DNA from tissue as from bacteria.

There are considerable biochemical differences between tissue and bacteria. Tissue contains multiple cells interconnected by proteins such as cell adhesion molecules, and the nuclei, which house the DNA and provide a further obstacle to isolating DNA. Bacteria, on the other hand, do not have the level of interconnection exhibited in tissue cells, and do not have nuclei. These differences, among others, are the reason for using more stringent conditions to isolate DNA from tissue, as opposed to bacteria.

Jaber states that GuHCL "inactives both RNase and DNase, dissociates nucleoproteins, and distrurbs cellular and subcellular structure, and its pH and ionic strength favour the native form of the DNA."[17] However, it continues to state that "[t]he effect of this solvent and the thermal degradation of the cellular organic material, combined with further protein denaturation by phenol and chloroform, have resulted in the isolation of biologically active and completely intact DNA from *M. tuberculosis* ...,"[18] not tissue. Moreover, this latter passage suggests that GuHCl may not be stringent enough to denature enough proteins to isolate DNA from tissue.

Thus, Jaber does not suggest that GuHCl is useful in isolating DNA from tissue, and the Office Action does not provide the requisite reason why one would reasonably expect that GuHCl would be useful in Chakrovarty's application.

A. Response to Office Action of April 8, 2009

The cited references use GuHCl and GITC as cell lysis reagents. In contrast, the present invention uses GuHCl to preserve mycobacteria and destroy the rest. Without a teaching the GuHCl is useful for this purpose, there would be no motivation to use GuHCl in the recited invention.

Jaber does not suggest that GuHCl would preserve mycobacteria while destroying the rest. Thus, there is no reason for one to reasonably predict that the recited

[17] Jaber at pate 579.

[18] Id.

solutions would be effective for their intended purpose. Accordingly, a *prima facie* case of obviousness has not been established.

Point III. The Examiner has not explained why one would combine the two references when the DNA sample in Chakravorty is isolated by high speed centrifugation while the DNA in Jaber is isolated by precipitation.

According to Jaber, once the bacteria is lysed, the sample is centrifuged at 10,000 g at 4°C for ten minutes. The resulting supernatant is thereafter transferred to a clean tube, and the DNA is precipitated with ice cold ethanol. According to Jaber, the ethanol precipitation step is crucial.[19] In contrast, Chakrovarty discloses a method of homogenizing tissue. The homongenate is centrifuged at 600 g for three minutes. The resulting supernatant is centrifuged at high speed, which results in the pelleting of the desired DNA sample.

In view of these differences in procedure, the Patent Office should explain why one would expect, nevertheless, that a DNA sample that is pure enough for PCR amplification can be obtained when using GuHCl.

A. Response to Office Action of April 8, 2009

In response to this argument, the Examiner contends that "an ordinary artisan would have been motivated to substitute known art-recognized equivalents, such as HuHCl and GITC, when preparing the buffers used in the method of Chakravorty."[20] However, as discussed above, GuHCl is not used as a cell lysis reagent. Instead, it is used to preserve the mycobacteria. Furthermore, unless there is some reason why one would expect GuHCl to be effective without centrifuging, a *prima facie* case of obviousness has not been established. Stating that GuHCl and GITC are art recognized substitutes for cell lysis reagents does not explain why one would reasonably expect that DNA extracted from cells lysed with GuHCl can be isolated by centrifugation instead of ethanol precipitation. Furthermore, it does not explain why one would have expected GUHCl to preserve the mycobacteria.

Without such an explanation, a *prima facie* case of obviousness has not been established.

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[19] Jaber at 579.

[20] Office Action at page 17.

Point IV. The Examiner has not explained why one would pick only GITC for GuHCl as opposed to substitute Jaber's entire lysis buffer formulation for Chakrovarty's lysis buffer.

Moreover, there is no reason provided why one would use only Jaber's GuHCl, instead of Jaber's entire lysis buffer. The lysis buffers taught by the individual references are different from each other; therefore, it is not as simple as substituting the GITC in Chakrovarty's buffer for the GuHCl used in Jaber's buffer (see Table 1). Specifically, Jaber discloses using GuHC1 and Tween 80, which are not used in Chakrovarty's lysis buffer. Furthermore, Jaber and Chakrovarty have different concentrations of EDTA and 2-mercaptoethanol.

Table 1: Side-by-Side Comparison of Buffers

Compound	Chakrovarty	Jaber	Recited Solution 1
GuHCL	0	6 M	3-6 M
GITC	5 M	0	
EDTA	25 mM	50 mM	20-30 mM
Tris-Cl	50 mM	0	40-60 mM
2-mercaptoethanol	0.2 M	1 mM	0.1-0.3 M
Sarcosyl	0.5 %	0	0.3-0.8 %
Tween 80	0	0.05 %	

In order to establish a *prima facie* case of obviousness, the references as a whole must be evaluated. Jaber does not commoditize GuHCl into an interchangeable part that can be used in other lysis buffers. Nor does Chakrovarty declare that GITC can be easily substituted for some other compound. There is no reason why one would believe that Jaber's GuHCl would be equally as effective in a lysis buffer without Tween 80, with half as much EDTA, or with 50 times less 2-mercaptoethanol. Without such a reason, a rejection under Section 103 cannot be maintained.

Furthermore, Jaber expressly states that phenol and chloroform are necessary to isolate intact DNA.[21] Chakrovarty does not require these compounds. Thus, without some reason why one would expect GuHCl to be effective without phenol or chloroform, there is no reason why one would combine Chakrovarty and Jaber.

[21] Jaber at 579.

A. Response to Office Action of April 8, 2009

The Examiner contends that, since GuHCl and GITC are art recognized substitutes, one would simply substitute one reagent for another instead without explaining why one would not instead substitute the entire buffer, or would reasonably believe that GuHCl would work equally as well within Jaber's and Chakrovarty's buffers. Without this explanation, a *prima facie* case of obviousness has not been established.

Accordingly, a *prima facie* case of obviousness has not been established.

II. REJECTION OF CLAIMS 127-129

Claims 127-129, which ultimately depend from claim 117, are patentable over these references for the same reason claim 117 is patentable over the combination of Chakravorty, Jaber and Herrnstadt.

Additionally, there is no motivation to combine these references. According to *Yamanouchi Pharmaceutical*, there must be some motivation for a hypothetical person of ordinary skill in the field at the time of the patent to take the myriad components of various teachings of prior art and combine them to create the claimed invention. *Yamanouchi Pharmaceutical Co. v. Danbury Pharmacal, Inc.*, 21 F.Supp.2d 366, 373, 48 U.S.P.Q.2d 1741 (S.D.N.Y. 1998), *aff'd*, 231 F.3d 1339 (Fed. Cir. 2000), *reh'g and reh'g en banc denied by*, 2000 U.S. App. LEXIS 34047 (Fed. Cir. 2000). In *Yamanouchi Pharmaceutical*, the court found that a skilled artisan would not be motivated to dispel all potential compounds disclosed in the cited reference, particularly when many of the disclosed compounds did not ever enter clinical trials due to insufficient potency, side effects or toxicity. *Id.*

Likewise, there is no motivation to pick over the vast number of possible primers to arrive at the particularly claimed primers. Without such a motivation, the references cannot be combined.

A. Response to Office Action of April 8, 2009

The Examiner contends that "an ordinary artisan would have been motivated to apply the teachings of Marchetti regarding dependence of PCR sensitivity on target length to the method resulting from the combined teachings of Chakravorty and Jabor." However, this does not explain why one reasonably expect that the recited primers would be useful.

The Examiner further contends that there are a finite number of primers disclosed in the GenBank reference.[22] The GenBank reference discloses a 1121 base pair sequence. In order to establish a *prima facie* case of obviousness, a reason by one would pick these specific primers must be established. With such a reason, this rejection should be withdrawn.

CONCLUSION

Accordingly, claim 117 is patentable over the cited references. Claims 118-120 and 124-129 are also patentable over the cited references by virtue of their dependence on claim 117. Therefore, in view of the amendments to the claims and remarks, Applicants respectfully request that the objections and rejections asserted in the Office Action of September 4, 2008 be reconsidered and withdrawn, that pending claims 117-120 and 124-129 be allowed. The Applicants further request that claims 130-132 be rejoined and allowed.

Respectfully submitted,

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[22] Office Action at page 19.